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## **Can genomic data alone tell us whether speciation happened with gene flow?**

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## **Abstract**

The allopatric model, which requires a period of geographical isolation for speciation to complete, has been the standard model in the modern era. Recently, “speciation with gene flow” has been widely discussed in relation to the model of “strict allopatry” and the level of DNA divergence across genomic regions. We wish to caution that genomic data by themselves may only permit the rejection of the simplest form of allopatry. Even a slightly more complex and realistic model that starts with sub-divided populations would be impossible to reject by the genomic data alone. To resolve this central issue of speciation, other forms of observations such as the sequencing of reproductive isolation genes or the identification of geographical barrier(s) will be necessary.

## **Text**

In the Modern Synthesis, speciation facilitated by allopatry (often referred to simply as allopatric speciation) has been the standard model for more than half a century (Mayr 1963; Coyne & Orr 2004). In this view, speciation requires a period of geographical isolation during which the diverging populations complete the process of speciation without experiencing gene flow. Although this period of strict allopatry is essential, events before and after this period may vary from species to species, potentially leading to false rejections of the allopatric model.

The conceptual basis for requiring a period of strict isolation is stated in Mayr (1963). Gene flow is perceived as a homogenizing force and even a tiny amount is assumed to be able to reverse the divergence. In this view, all genic regions gradually evolve functional characteristics that are cohesive within the same species but each region would have negative interactions with the genome of the diverging population. While epistasis in fitness is commonly accepted in the Modern Synthesis (Dobzhansky 1937; Wright 1941), strict allopatry postulates genome-wide “cohesiveness” (Wu 2001).

In the modern view of genetics, many large segments of the genome may harbor no loci of fitness consequence between nascent species (Wu & Ting 2004; Poelstra *et al.* 2014; Fontaine *et al.* 2015; Malinsky *et al.* 2015; Zhang *et al.* 2016). These neutral gene regions can easily travel between nascent species during speciation while selection operates only on genes that have diverged functionally. In this genic view (Feder *et al.* 2012; Seehausen *et al.* 2014), gene flow would not retard ecological divergence when different alleles are selected against in different environments. The implication is that gene flow would pose little problem for species divergence and strict allopatry is not necessarily the dominant mode of speciation (Schilthuizen 2000; Schluter 2001; Schluter & Conte 2009; Feder *et al.* 2013).

Despite increasing discussions of this alternative model in the era of genomics, strict allopatry may still be necessary if reproductive isolation is taken into account. Consider the simple Dobzhansky-Muller model of hybrid incompatibility where the initial state is a two locus (a, b)

haplotype. Let  $a$  and  $b$  evolve toward  $A$  and  $B$ , respectively, under positive selection but  $A$  and  $B$  are incompatible. In full geographical isolation, population 1 may evolve to  $(A, b)$  while population 2 evolves to  $(a, B)$  and hybrid incompatibility would ensue. If gene flow occurs between the two diverging populations, the mutual interference between  $a \rightarrow A$  and  $b \rightarrow B$  would render each pathway impassable, effectively stopping the evolution of postmating isolation. These dynamics, first explored in the context of gene silencing after gene duplication, are highly stochastic (Li 1980; Maruyama & Takahata 1981; Watterson 1983). Although the same dynamics have been analyzed for the evolution of hybrid incompatibility only in deterministic terms (Agrawal *et al.* 2011; Nosil & Flaxman 2011; Bank *et al.* 2012), it is clear that gene flow can strongly retard the evolution of reproductive incompatibility.

Therefore, depending on the nature of genic interactions, gene flow can be seen either as a neutral force or as a strong retardant of speciation. “Strict allopatry” vs. “speciation with gene flow” obviously must be resolved by empirical observations (Sousa & Hey 2013; Faria *et al.* 2014; Seehausen *et al.* 2014). It has been optimistically suggested that the torrent of genomic data will provide a clear answer. In this short note, we raise caveats against this optimism. It is unlikely that the issue of “speciation with gene flow” will be resolved by genomic data alone, regardless of the sophistication of the statistical tools (Becquet & Przeworski 2009; Strasburg & Rieseberg 2010, 2011, 2013; Sousa *et al.* 2011) or the amount of genomic data. Other types of observations will be needed in conjunction with the genomic data.

In previous tests of strict allopatry, the model is the Level 1 model of allopatry shown in Fig. 1a. An uninterrupted period of geographical isolation, during which gene flow ceases, is portrayed between  $T_1$  and  $T_2$ . Speciation is completed by  $T_2$  after which the species may come into contact again. Tests of allopatry are usually based on this Level 1 model because even a slightly more complex model will be nearly impossible to reject, if it is testable at all, by the genomic data alone (see Fig. 1b and text below) (Won *et al.* 2005; Geraldts *et al.* 2006, 2011; Niemiller *et al.* 2008; Nadachowska & Babik 2009; Ellegren *et al.* 2012; Garrigan *et al.* 2012; Runemark *et al.* 2012; Mailund *et al.* 2012; Osborne *et al.* 2013; Sousa *et al.* 2013; Winker *et al.* 2013; Kronforst *et al.* 2013; Leaché *et al.* 2013; Muñoz *et al.* 2013; Herrig *et al.* 2014).

In statistical terms, the Level 1 model (Fig. 1a) is an excellent null hypothesis that assumes that all genomic regions have diverged for the same length of time. In contrast, speciation with gene flow would result in some loci, especially those pertaining to species divergence (or “speciation genes”, for short), becoming more divergent than others. The rationale for testing such a simple model is that the failure to reject it may also mean that strict allopatry in general cannot be rejected (Innan & Watanabe 2006; Presgraves & Yi 2009; Yamamichi *et al.* 2012). This is because complex models with more parameters would be even more difficult to reject. Importantly, given massive genomic data and refined statistical methods, there should be sufficient statistical power to reject the null model if it is indeed incorrect.

The issue arises because the null model of Fig. 1a is often rejected (Osada & Wu 2005; Pinho & Hey 2010; Garrigan *et al.* 2012; Kronforst *et al.* 2013; Leaché *et al.* 2013; Muñoz *et al.* 2013; Osborne *et al.* 2013). Highly divergent regions, in the form of “genomic islands” that are not expected by the null model have been widely reported (Ellegren *et al.* 2012; Carneiro *et al.* 2014; Poelstra *et al.* 2014; Malinsky *et al.* 2015; Toews *et al.* 2016; but see Renaut *et al.* 2013; Pennisi 2014; Burri *et al.* 2015). Although some results may have been mis-interpreted due to low polymorphism instead of high divergence (Cruickshank & Hahn 2014; Payseur & Rieseberg 2016; Wolf & Ellegren 2017), many are likely correct in rejecting the null model of Fig. 1a.

The rejection of Level 1 allopatry thus raises the bar - “Can a more realistic model of allopatric speciation be rejected?” Fig. 1b shows a Level 2 model of allopatric speciation. Unlike in the Level 1 model whereby geographical isolation is imposed on a geographically panmictic population, full isolation in nature may often happen between partially-differentiated geographical populations. Indeed, many studies following the Takahata *et al.* (1995) analysis have found the ancestral population to have an effective size much larger than the extant one (Satta *et al.* 2004; Osada & Wu 2005; Won & Hey 2005; Zhou *et al.* 2007; Mailund *et al.* 2014). A plausible explanation for the very large effective size would be populations with deep structure (Osada & Wu 2005; Zhou *et al.* 2007; Becquet & Przeworski 2009), hence supporting the model of Fig. 1b.

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If allopatry is indeed the most common mode of speciation and generally follows the Level 2 model, genomic data could not distinguish between “strict allopatry” and “speciation with gene flow”. Note that genomic data by themselves can only inform whether there has been gene flow between  $T_0$  (when the species started to differentiate) and  $T_2$  (when speciation was completed). They cannot tell us whether there exists a period ( $T_1$  to  $T_2$ ) without gene flow. Unless the pattern of geographical differentiation at  $T_1$  is known, it will not be possible to test the Level 2 model of Fig. 1b. Of course, methods exist for inferring the ancient population structure but such models (Takahata *et al.* 1995; Takahata & Satta 1997; Yang 2002; Innan & Watanabe 2006) have to assume a period of strict allopatry, akin to the Level 2 model. In other words, either allopatry is true or the ancient population structure has to be known in order to test the Level 2 model. Genomic data alone are therefore insufficient to reject such a model.

Not knowing the ancient population structure raises other conceptual issues. It has been suggested that population differentiation between  $T_0$  and  $T_1$  in Fig. 1b should be considered “incipient speciation”, implying that the Level 2 model is also a model of “speciation with gene flow” (Butlin 2008, 2010). As stated earlier, the models of allopatric speciation require a period of strict isolation with no gene flow ( $T_1 - T_2$  in Fig. 1). Thus, while the rejection of allopatry entails the proof that such a period is absent, other events before  $T_1$ , or after  $T_2$ , are peripheral to the allopatry debate. Furthermore, it has been demonstrated that when the differentiation at  $T_1$  has little to do with incipient speciation (e.g., in simulations of neutral divergence), allopatric speciation simulated by the Level 2 model could still be interpreted as “speciation with gene flow” (Pinho & Hey 2010). To take the simulation

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approach a step further, we suggest a prospective approach starting with genomic data of actual geographical populations (such as the extant human populations). We will then be able to test whether the simulated new species, when analyzed retrospectively, would falsely reject the model of allopatry. The approach is based on realistic extant populations and should be a useful tool for analyzing various allopatric models of speciation.

More complex models of allopatry (beyond Level 2) have been applied to many taxa (Duvaux *et al.* 2011; Nadachowska-Brzyska *et al.* 2013; Roux *et al.* 2013; Tine *et al.* 2014; Lohse *et al.* 2015; Filatov *et al.* 2016; Le Gac *et al.* 2016; Christe *et al.* 2017). These are usually models with extensive recent gene flow applied to “species pairs” that have not developed full reproductive isolation (RI). For these taxa, the issue is not gene flow. Instead, it is whether the taxa will need a long period of strict allopatry in the future to develop full RI. (One may also ask, retrospectively, whether the absence of full RI might be connected to insufficiently long period of geographical isolation in the past.)

It seems clear that information in addition to genomic data will be necessary for testing the more realistic models of allopatric speciation. There are at least two options. First, we may use functional genomic means to test the general allopatric models. In allopatry, the divergence is a function of the timing of erecting the geographical isolation and “speciation genes” underlying reproductive isolation should be no more divergent than the rest of the genome. In speciation with gene flow, true speciation genes that evolve in the very incipient stage should be older than the others (Ting *et al.* 2001).

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Second, we suggest that the bio-geographical records of species in question be incorporated into the analysis of genomic data. It would seem desirable to analyze species pairs that are separated by a well-defined geographical barrier as the geological record of the barrier can guide tests of gene flow with timing information. The Isthmus of Panama is the best-known example. Its gradual and irreversible closure has made speciation between the Pacific and Atlantic sides the best examples of allopatry (Palumbi 1994; Bacon *et al.* 2015). Besides permanent barriers, non-permanent barriers exist in many terrestrial, aquatic and marine environments. Speciation associated with such barriers may be informative about the roles of gene flow. The Indo-Pacific Barrier near the Strait of Malacca could be such an example. In addition, a non-permanent barrier may continue to produce new species, hence providing valuable data on speciation events of different ages.

Whether speciation can proceed in the presence of gene flow is important in many aspects of biology including genetics, development, behavior, ecology and biogeography. On one hand, it may seem difficult to envisage the emergence of thousands of species of beetles in the Amazon, all requiring periods of strict allopatry (Erwin 1996; Hoorn *et al.* 2010). On the other hand, the model of “speciation with gene flow” will require the convincing rejection of strict allopatry, which remains challenging even with the massive genomic data.

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## References

- Agrawal AF, Feder JL, Nosil P (2011) Ecological divergence and the origins of intrinsic postmating isolation with gene flow. *International Journal of Ecology*, **2011**.
- Bacon CD, Silvestro D, Jaramillo C *et al.* (2015) Biological evidence supports an early and complex emergence of the Isthmus of Panama. *Proceedings of the National Academy of Sciences*, **112**, 6110–6115.
- Bank C, Bürger R, Hermisson J (2012) The limits to parapatric speciation: Dobzhansky–Muller incompatibilities in a continent–island model. *Genetics*, **191**, 845–863.
- Becquet C, Przeworski M (2009) Learning about modes of speciation by computational approaches. *Evolution*, **63**, 2547–2562.
- Burri R, Nater A, Kawakami T *et al.* (2015) Linked selection and recombination rate variation drive the evolution of the genomic landscape of differentiation across the speciation continuum of *Ficedula* flycatchers. *Genome research*, **25**, 1656–1665.
- Butlin RK (2008) Population genomics and speciation. , **2008**.
- Butlin RK (2010) Population genomics and speciation. *Genetica*, **138**, 409–418.
- Carneiro M, Albert FW, Afonso S *et al.* (2014) The genomic architecture of population divergence between subspecies of the European Rabbit. *PLoS Genetics*, **10**, e1003519.

- Christe C, Stolting KN, Paris M *et al.* (2017) Adaptive evolution and segregating load contribute to the genomic landscape of divergence in two tree species connected by episodic gene flow. *Molecular Ecology*, **26**, 59–76.
- Coyne JA, Orr HA (2004) *Speciation*. Sinauer Associates Sunderland, MA.
- Cruickshank TE, Hahn MW (2014) Reanalysis suggests that genomic islands of speciation are due to reduced diversity, not reduced gene flow. *Molecular ecology*, **23**, 3133–3157.
- Dobzhansky T (1937) *Genetics and the Origin of Species*. Columbia University Press.
- Duvaux L, Belkhir K, Boulesteix M, Boursot P (2011) Isolation and gene flow: Inferring the speciation history of European house mice. *Molecular Ecology*, **20**, 5248–5264.
- Ellegren H, Smeds L, Burri R *et al.* (2012) The genomic landscape of species divergence in Ficedula flycatchers. *Nature*, **491**, 756–760.
- Erwin TL (1996) Biodiversity at its utmost: tropical forest beetles. *Biodiversity II. Understanding and protecting our biological resources*. Joseph Henry Press, Washington, 27–40.
- Faria R, Renaut S, Galindo J *et al.* (2014) Advances in Ecological Speciation: An integrative approach. *Molecular Ecology*, **23**, 513–521.
- Feder JL, Egan SP, Nosil P (2012) The genomics of speciation-with-gene-flow. *Trends in Genetics*, **28**, 342–350.
- Feder JL, Flaxman SM, Egan SP, Comeault AA, Nosil P (2013) Geographic mode of speciation and genomic divergence. *Annual Review of Ecology, Evolution, and Systematics*, **44**, 73–97.
- Filatov DA, Osborne OG, Papadopulos AST (2016) Demographic history of speciation in a Senecio altitudinal hybrid zone on Mt. Etna. *Molecular Ecology*, **25**, 2467–2481.
- Fontaine MC, Pease JB, Steele A *et al.* (2015) Extensive introgression in a malaria vector species complex revealed by phylogenomics. *Science*, **347**, 1258524.
- Le Gac M, Metegnier G, Chomérat N *et al.* (2016) Evolutionary processes and cellular functions underlying divergence in *Alexandrium minutum*. *Molecular Ecology*, **25**, 5129–5143.
- Garrigan D, Kingan SB, Geneva AJ *et al.* (2012) Genome sequencing reveals complex speciation in the *Drosophila simulans* clade. *Genome Research*, **22**, 1499–1511.

- Geraldes A, Basset P, Smith KL, Nachman MW (2011) Higher differentiation among subspecies of the house mouse (*Mus musculus*) in genomic regions with low recombination. *Molecular Ecology*, **20**, 4722–4736.
- Geraldes A, Ferrand N, Nachman MW (2006) Contrasting patterns of introgression at X-linked loci across the hybrid zone between subspecies of the European rabbit (*Oryctolagus cuniculus*). *Genetics*, **173**, 919–933.
- Herrig DK, Modrick AJ, Brud E, Llopart A (2014) Introgression in the *Drosophila subobscura*-*D. madeirensis* sister species: Evidence of gene flow in nuclear genes despite mitochondrial differentiation. *Evolution*, **68**, 705–719.
- Horn C, Wesselingh FP, Ter Steege H *et al.* (2010) Amazonia through time: Andean uplift, climate change, landscape evolution, and biodiversity. *science*, **330**, 927–931.
- Innan H, Watanabe H (2006) The effect of gene flow on the coalescent time in the human-chimpanzee ancestral population. *Molecular Biology and Evolution*, **23**, 1040–1047.
- Kronforst MR, Hansen MEB, Crawford NG *et al.* (2013) Hybridization Reveals the Evolving Genomic Architecture of Speciation. *Cell Reports*, **5**, 666–677.
- Leaché AD, Harris RB, Maliska ME, Linkem CW (2013) Comparative species divergence across eight triplets of spiny lizards (*Sceloporus*) using genomic sequence data. *Genome Biology and Evolution*, **5**, 2410–2419.
- Li W-H (1980) Rate of gene silencing at duplicate loci: a theoretical study and interpretation of data from tetraploid fishes. *Genetics*, **95**, 237–258.
- Lohse K, Clarke M, Ritchie MG, Etges WJ (2015) Genome-wide tests for introgression between cactophilic *Drosophila* implicate a role of inversions during speciation. *Evolution*, **69**, 1178–1190.
- Mailund T, Halager AE, Westergaard M *et al.* (2012) A New Isolation with Migration Model along Complete Genomes Infers Very Different Divergence Processes among Closely Related Great Ape Species. *PLoS Genetics*, **8**, e1003125.
- Mailund T, Munch K, Schierup MH (2014) Lineage sorting in apes. *Annual Review of Genetics*, **48**, 519–535.
- Malinsky M, Challis RJ, Tyers AM *et al.* (2015) Genomic islands of speciation separate cichlid ecomorphs in an East African crater lake. *Science*, **350**, 1493–1498.

- Maruyama T, Takahata N (1981) Numerical studies of the frequency trajectories in the process of fixation of null genes at duplicated loci. *Heredity*, **46**, 49–57.
- Mayr E (1963) *Animal species and evolution*. Belknap Press of Harvard University Press Cambridge, Massachusetts.
- Muñoz MM, Crawford NG, McGreevy TJ *et al.* (2013) Divergence in coloration and ecological speciation in the *Anolis marmoratus* species complex. *Molecular Ecology*, **22**, 2668–2682.
- Nadachowska-Brzyska K, Burri R, Olason PI *et al.* (2013) Demographic Divergence History of Pied Flycatcher and Collared Flycatcher Inferred from Whole-Genome Re-sequencing Data. *PLoS Genetics*, **9**, e1003942.
- Nadachowska K, Babik W (2009) Divergence in the face of gene flow: The case of two newts (Amphibia: Salamandridae). *Molecular Biology and Evolution*, **26**, 829–841.
- Niemiller ML, Fitzpatrick BM, Miller BT (2008) Recent divergence with gene flow in Tennessee cave salamanders (Plethodontidae: Gyrinophilus) inferred from gene genealogies. *Molecular Ecology*, **17**, 2258–2275.
- Nosil P, Flaxman SM (2011) Conditions for mutation-order speciation. *Proceedings of the Royal Society B-Biological Sciences*, **278**, 399–407.
- Osada N, Wu C-I (2005) Inferring the mode of speciation from genomic data. *Genetics*, **169**, 259–264.
- Osborne OG, Batstone TE, Hiscock SJ, Filatov DA (2013) Rapid speciation with gene flow following the formation of Mt. Etna. *Genome Biology and Evolution*, **5**, 1704–1715.
- Palumbi SR (1994) Genetic divergence, reproductive isolation, and marine speciation. *Annual review of ecology and systematics*, 547–572.
- Payseur BA, Rieseberg LH (2016) A genomic perspective on hybridization and speciation. *Molecular ecology*, **25**, 2337–2360.
- Pennisi E (2014) Disputed islands. *Science*, **345**, 611–613.
- Pinho C, Hey J (2010) Divergence with gene flow: models and data. *Annual review of ecology, evolution, and systematics*, **41**, 215–230.
- Poelstra JW, Vijay N, Bossu CM *et al.* (2014) The genomic landscape underlying phenotypic integrity in the face of gene flow in crows. *Science*, **344**, 1410–1414.

Presgraves DC, Yi S V. (2009) Doubts about complex speciation between humans and chimpanzees. *Trends in Ecology and Evolution*, **24**, 533–540.

Renaut S, Grassa CJ, Yeaman S *et al.* (2013) Genomic islands of divergence are not affected by geography of speciation in sunflowers. *Nature communications*, **4**, 1827.

Roux C, Tsagkogeorga G, Bierne N, Galtier N (2013) Crossing the species barrier: Genomic hotspots of introgression between two highly divergent *Drosophila* species. *Molecular Biology and Evolution*, **30**, 1574–1587.

Runemark A, Hey J, Hansson B, Svensson EI (2012) Vicariance divergence and gene flow among islet populations of an endemic lizard. *Molecular Ecology*, **21**, 117–129.

Satta Y, Hickerson M, Watanabe H, O’Higin C, Klein J (2004) Ancestral population sizes and species divergence times in the primate lineage on the basis of intron and BAC end sequences. *Journal of molecular evolution*, **59**, 478–487.

Schilthuizen M (2000) Dualism and conflicts in understanding speciation. *BioEssays*, **22**, 1134–1141.

Schluter D (2001) Ecology and the origin of species. *Trends in ecology & evolution*, **16**, 372–380.

Schluter D, Conte GL (2009) Genetics and ecological speciation. *Proceedings of the National Academy of Sciences*, **106**, 9955–9962.

Seehausen O, Butlin RK, Keller I *et al.* (2014) Genomics and the origin of species. *Nature Reviews Genetics*, **15**, 176–192.

Sousa VC, Carneiro M, Ferrand N, Hey J (2013) Identifying loci under selection against gene flow in isolation-with-migration models. *Genetics*, **194**, 211–233.

Sousa VC, Grelaud A, Hey J (2011) On the nonidentifiability of migration time estimates in isolation with migration models. *Molecular ecology*, **20**, 3956–3962.

Sousa V, Hey J (2013) Understanding the origin of species with genome-scale data: modelling gene flow. *Nat Rev Genet*, **14**.

Strasburg JL, Rieseberg LH (2010) How robust are “isolation with migration” analyses to violations of the im model? A simulation study. *Molecular Biology and Evolution*, **27**, 297–310.

Strasburg JL, Rieseberg LH (2011) Interpreting the estimated timing of migration events between hybridizing species. *Molecular Ecology*, **20**, 2353–2366.

- Accepted Article
- Strasburg JL, Rieseberg LH (2013) Methodological challenges to realizing the potential of hybridization research. *Journal of Evolutionary Biology*, **26**, 259–260.
- Takahata N, Satta Y (1997) Evolution of the primate lineage leading to modern humans: phylogenetic and demographic inferences from DNA sequences. *Proceedings of the National Academy of Sciences*, **94**, 4811–4815.
- Takahata N, Satta Y, Klein J (1995) Divergence time and population size in the lineage leading to modern humans. *Theoretical population biology*, **48**, 198–221.
- Tine M, Kuhl H, Gagnaire P-A *et al.* (2014) European sea bass genome and its variation provide insights into adaptation to euryhalinity and speciation. *Nature Communications*, **5**, 5770.
- Ting C-T, Takahashi A, Wu C-I (2001) Incipient speciation by sexual isolation in *Drosophila*: concurrent evolution at multiple loci. *Proceedings of the National Academy of Sciences*, **98**, 6709–6713.
- Toews DPL, Taylor SA, Vallender R *et al.* (2016) Plumage Genes and Little Else Distinguish the Genomes of Hybridizing Warblers. *Current Biology*, **26**, 2313–2318.
- Watterson GA (1983) On the time for gene silencing at duplicate loci. *Genetics*, **105**, 745–766.
- Winker K, McCracken KG, Gibson DD, Peters JL (2013) Heteropatric speciation in a duck, *Anas crecca*. *Molecular Ecology*, **22**, 5922–5935.
- Wolf JBW, Ellegren H (2017) Making sense of genomic islands of differentiation in light of speciation. *Nature Reviews Genetics*, **18**, 87–100.
- Won YJ, Hey J (2005) Divergence population genetics of chimpanzees. *Molecular Biology and Evolution*, **22**, 297–307.
- Won Y-J, Sivasundar A, Wang Y, Hey J (2005) On the origin of Lake Malawi cichlid species: a population genetic analysis of divergence. *Proceedings of the National Academy of Sciences of the United States of America*, **102**, 6581–6586.
- Wright S (1941) The statistical consequences of Mendelian heredity in relation- to speciation. In “The new systematics,” ed. by J. Huxley.
- Wu C (2001) The genic view of the process of speciation. *Journal of Evolutionary Biology*, **14**, 851–865.
- Wu C-I, Ting C-T (2004) Genes and speciation. *Nature Reviews Genetics*, **5**, 114–122.

Accepted Article

Yamamichi M, Gojobori J, Innan H (2012) An autosomal analysis gives no genetic evidence for complex speciation of humans and chimpanzees. *Molecular Biology and Evolution*, **29**, 145–156.

Yang Z (2002) Likelihood and Bayes estimation of ancestral population sizes in hominoids using data from multiple loci. *Genetics*, **162**, 1811–1823.

Zhang W, Dasmahapatra KK, Mallet J, Moreira GRP, Kronforst MR (2016) Genome-wide introgression among distantly related *Heliconius* butterfly species. *Genome biology*, **17**, 17:25.

Zhou R, Zeng K, Wu W *et al.* (2007) Population genetics of speciation in nonmodel organisms: I. Ancestral polymorphism in mangroves. *Molecular Biology and Evolution*, **24**, 2746–2754.

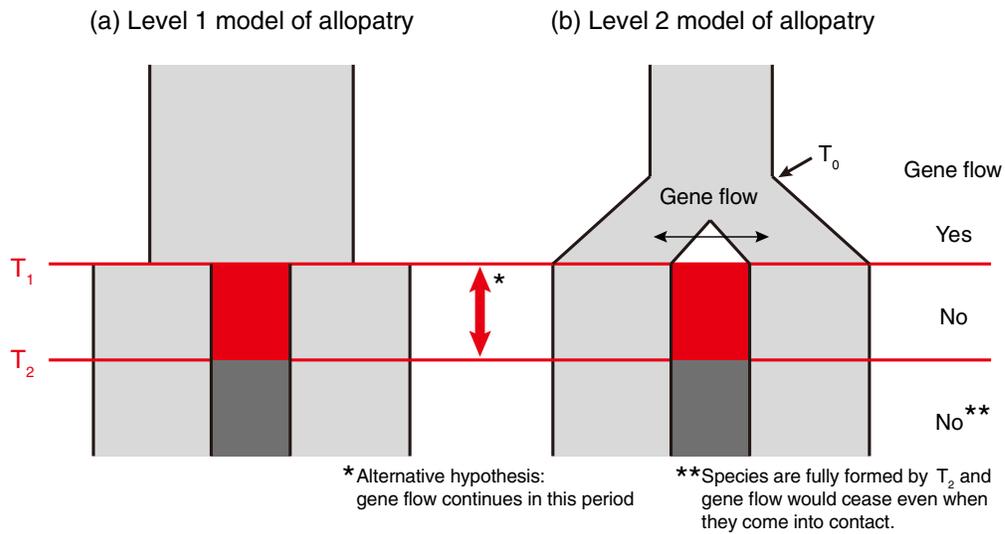
#### **Data Accessibility**

No data needed in the main text.

#### **Author Contributions**

C-I.W. wrote the paper with input from M.Y. Edited by all authors.

Figures



**Fig. 1** Two levels of models of allopatric speciation.  $T_1 - T_2$  is the period of strict geographical isolation with no gene flow that is required for the completion of allopatric speciation. The red arrow with \* indicates the alternative model with gene flow. In the subsequent period ( $T_2 - \text{present}$ ), geographical isolation is no longer necessary. (a) Level 1 model of allopatry – Geographical isolation is imposed on a single undivided population. (b) Level 2 model of allopatry – In the time period  $T_0 - T_1$ , a species is first divided into two geographical populations. The model considers only populations that have not diverged in traits and genes germane to speciation. In other words, when geographical isolation is imposed at  $T_1$ , the geographical populations do not show characters of incipient speciation.